teams of directionally-similar motors, i.e., kinesin-1 and kinesin-2, facilitate long range cargo transport in a complex intracellular environment.

674-Pos Board B454
Cooperative Transport by Populations of Fast and Slow Kinesins Uncovers Novel Family-Dependent Motor Characteristics Important for in vivo Function
Göker Arpağ[1], Shankar Shastry[2], William O. Hancock[3], **Erkan Tüzel**[1].
[1]Department of Physics, Worcester Polytechnic Institute, Worcester, MA, USA, [2]Department of Biomedical Engineering, The Pennsylvania State University, University Park, PA, USA, [3]Department of Biomedical Engineering, Pennsylvanie State University, University Park, PA, USA.
Intracellular cargo transport frequently involves multiple motor types, either having opposite directionality or having the same directionality but different speeds. Although significant progress has been made in characterizing kinesin motors at the single-molecule level, predicting their ensemble behavior is challenging and requires tight coupling between experiments and modeling to uncover the underlying motor behavior. To understand how diverse kinesins attached to the same cargo coordinate their movement, we carried out microtubule gliding assays using pairwise mixtures of motors from the kinesin-1, 2, 3, 5 and 7 families engineered to have identical run lengths and surface attachments. Uniform motor densities were used and microtubule gliding speeds were measured for varying proportions of fast and slow motors. A coarse-grained computational model of gliding assays was developed and found to recapitulate the experiments. The simulations show that the force-dependence of detachment is the key parameter that determines gliding speed in multi-motor assays and provide estimates for force-dependent dissociation rates suggesting that kinesin-1 and the mitotic motors kinesin-5 and −7 maintain microtubule association against loads, while kinesin-2 and −3 readily detach. Using these predictions, we are investigating how these motors carry scaffold proteins and quantum dot cargo in teams to carry out distinct mechanical tasks in cells. Our work uncovers unexpected motor behavior in multi-motor ensembles and clarifies functional differences between kinesins.

675-Pos Board B455
Kinesin Regulation Dynamics through Cargo Delivery, a Single Molecule Investigation
Anthony P.I. Kovacs[1], Jonathan M. Kessler[1], Huawen Lin[2], Susan K. Dutcher[2], Yan Mei Wang[1].
[1]Physics, Washington University in St. Louis, Saint Louis, MO, USA, [2]Genetics, Washington University School of Medicine, Saint Louis, MO, USA.
Kinesins are microtubule-based motors that deliver cargo to their destinations in a highly regulated manner. Although numerous regulators of cargo delivery have been identified in recent years, the regulation mechanism for kinesin through the cargo delivery and recycling process is not known. By performing single molecule fluorescence imaging measurements in Chlamydomonas flagella, which are 200 nm in diameter and 10 microns in length with 9 sets of doublet microtubules, we tracked intraflagellar transport (IFT) trains, BBSome cargoes, and kinesin-2 motors through the entire cargo delivery process and determined their reorganization and recycling dynamics. Upon arrival at the microtubule plus end at the flagellar tip, (1) IFT trains and BBSome cargoes remain intact and dissociate away from the kinesins and microtubules together, diffuse, and reorganize along the flagellar membrane over the course of 2.3 seconds before commencing retrograde travel. (2) Kinesin motors remain bound to and diffuse along microtubules for 1.3 seconds before dissociating into the flagellar lumen for recycling.

676-Pos Board B456
Sensitivity of Multiple-Kinesin Transport to Microtubule Lattice Defects
K.M.Rifat Faysal[1], Stephen J. King[2], **Jing Xu**[1].
[1]Physics Graduate Group, University of California, Merced, Merced, CA, USA, [2]Burnett School of Biomedical Sciences, University of Central Florida, Orlando, FL, USA.
Microtubules are fundamental biopolymers in cells, formed via self-assembly of tubulin dimers. Defects in microtubule lattices have been observed, including point defects (missing tubulin dimers) and line defects (protofilament disruption). Microtubule-based molecular motors enable long-range transport in cells. Potential impact of microtubule lattice defects on intracellular transport is not yet understood. Here we vary microtubule polymerization conditions to uncontrollably tune defect presence in microtubule lattices, and use single-molecule-type optical trapping experiments to investigate the impact of such defect on multiple-kinesin transport. We find that kinesin-based cargoes pause preferentially at specific locations along individual microtubules, and that the pause frequency and duration increase with increasing presence of defects in microtubules. Additionally, we find that the dissociation rates of multiple-kinesin-based cargoes are also strongly dependent on the specific microtubules they travel along. Taken together, our study highlights a previously unexplored, and important role of microtubule lattice assembly in controlling intracellular transport.

677-Pos Board B457
Characterizing the Allosteric Effect of Nucleotide and Inhibitor Binding to the Kinesin Motor Domain
Guido Scarabelli, Barry J. Grant.
The University of Michigan, Ann Arbor, MI, USA.
Kinesin motor domains couple cycles of ATP hydrolysis to cycles of microtubule binding and conformational changes that result in directional force and movement on microtubules. General principles of this mechanochemical coupling have now been established. However, fundamental atomistic details of the underlying allosteric mechanisms remain unknown. This lack of knowledge hampers the development of new inhibitors and limits our understanding of how disease-associated mutations in distal sites can interfere with the fidelity of motor domain function. Here we combine unbiased molecular dynamics simulations, bioinformatics analysis and in-silico mutational studies to elucidate the structural dynamic effects of nucleotide turnover and allosteric inhibition. Multiple replica simulations of kinesin-1 and kinesin-5 bound to ATP-, ADP- and kinesin-5 specific inhibitors were used to create residue-based networks that characterize the internal dynamic coordination of functional regions. This analysis predicts the intervening residues involved in the dynamic coupling of nucleotide, microtubule, neck-linker and inhibitor binding sites. Regions identified include the nucleotide binding switch regions, loop5, loop7, α4-α5-loop13, α1 and β4-β6-β7. Also evident were nucleotide and inhibitor dependent shifts in the dynamic coupling paths linking functional sites. In particular, inhibitor binding to the loop5 region affects β-sheet residues and α1 leading to a dynamic decoupling of nucleotide, microtubule and neck-linker binding sites. Additional analysis of point mutations in kinesin-5, including P131 (loop5), Q78/I79 (α1), E166 (loop7), K272/I273 (β7) G325/G326 (loop13), support their predicted role in mediating the dynamic coupling of distal functional surfaces. Collectively, our results and approach provide a framework for explaining how binding events and point mutations can alter dynamic couplings critical for kinesin motor domain function.

678-Pos Board B458
Effect of Force and Discrete Step-Size on the Velocity Distribution of Processive Molecular Motors
Huong T. Vu[1], Shaon Chakrabarti[1], Michael Hinczewski[2], D. Thirumalai[1].
[1]Biophysics, IPST, University of Maryland, Hyattsville, MD, USA, [2]Department of Physics, Case Western Reserve University, Cleverland, OH, USA.
Understanding the distribution of velocities of identical motors as they walk along polar tracks is important in deciphering their locomotion mechanism. We created a three parameter kinetic model to describe the velocity, $P(v)$, and run-length distributions of generic molecular motors that step (forward and backward) on a track and have finite processivity. We obtain exact analytic results for the run-length and velocity distributions as a function of external resistive force. Remarkably, our theory fits Kinesin-1 data very well at zero force, and reproduces measured stall-force and ratio of the stepping rates. We extend the model to predict the behavior of Kinesin-1 velocity distributions under load. When extended to non-zero loads, the theory makes two interesting predictions. One is that the $P(v)$ is non-Gaussian and the other is a bimodal structure in $P(v)$. The bimodal structure, a feature that remains even with a finite step-size distribution, is a direct consequence of the discrete step-size of Kinesin-1. Although we analyze only Kinesin-1 data, our results are general and should hold for any processive motor, which walks on a polar track with a discrete step-size.

679-Pos Board B459
Development of Photo-Controlled Molecular Shuttle Utilizing ATP Driven Motor Kinesin
Naozumi Numata[1], Kazunori Kondo[2], Shinsaku Maruta[1,2].
[1]Div. Bioinfo., Grad. Sch Eng., Soka Univ., Tokyo, Japan, [2]Dept. Bioinfo., Fac. Eng., Soka Univ., Tokyo, Japan.
The ATP driven motor kinesin has many possibilities for the application to the molecular machines. Previously, we introduced photochromic molecules into the functional key region of the kinesin as a photo-switch and tried to control the function of kinesin by ultraviolet (UV) and visible (VIS) light irradiations. The kinesin mutant S275C modified with thiol reactive azobenzene derivative exhibited photo-controlled ATPase activity correlating to photo isomerization of photochromic molecules.
In this study, we tried to prepare the photo-controlled molecular shuttles using kinesin and liposome modified with photochromic molecule.